Amendments to the Claims

Claim 1 (currently amended): A matched set of fluorescent dyes comprising-at least two or more different fluorescent dyes of formula (I):

(I)

wherein n is different for each said dye and is 1, 2, or 3;

 Z^1 and Z^2 independently represent the carbon atoms necessary to complete a phenyl or naphthyl ring system;

one of groups R¹ and R² is the group:

$$-(CH_2)_p$$
 $--C-NH-(CH_2)_q$ $--Y$

where Y is a target bonding group;

remaining group R^1 or R^2 is selected from $-(CH_2)_4-W$ or $-(CH_2)_r-H$;

group R^3 is hydrogen, except when either R^1 or R^2 is $-(CH_2)_r-H$, in which case R^3 is W;

W is selected from sulphonic acid and sulphonate;

p is an integer from 3 to 6;

q is selected to be 2 or 3; and

r is an integer from 1 to 5;

and their salts thereof;

characterised in that and further wherein when n of two of said dyes differs by +1, one of p, q and r of said two dyes differs by -1.

Claim 2 (currently amended): A matched set of fluorescent dyes comprising at least two different fluorescent dyes of formula (II):

$$\mathbb{R}^3$$

(II)

wherein n is different for each said dye and is 1, 2, or 3; one of groups R^1 and R^2 is the group:

$$-(CH_2)_p$$
 $--(CH_2)_q$ $--Y$

where Y is a target bonding group;

remaining group R^1 or R^2 is selected from $-(CH_2)_4-W$ or $-(CH_2)_r-H$;

group R^3 is hydrogen, except when either R^1 or R^2 is $-(CH_2)_r-H$, in which case R^3 is W;

W is selected from sulphonic acid and sulphonate;

p is an integer from 3 to 6;

q is selected to be 2 or 3; and

r is an integer from 1 to 5;

and their salts thereof;

eharacterised in that and further wherein when n of two of said dyes differs by +1, one of p, q and r of said two dyes differs by -1.

Claim 3 (currently amended): A-The matched set according to of dyes of claim 1 or claim 2 comprising at least two different fluorescent dyes wherein according to formula (I) or (II) in which:

n is selected to be 1 or 2;

p is selected to be 4 or 5;

q is selected to be 2 or 3; and

r is-selected to be 1, 2 or 3.

Claim 4 (currently amended): A matched set according to any of claims 1 to 3-The matched set of dyes of claim 1 or claim 2, wherein said target bonding group Y in each dye of the set of dyes is the same and is selected from a the group consisting of maleimido group groups and an iodoacetamido groups group.

Claim 5 (currently amended): A matched set according to The matched set of dyes of claim 4 wherein in each said dye Y is a maleimido group.

Claim 6 (currently amended): A matched set according to any of claims 1 to 5-The matched set of dyes of claim 1 or claim 2, wherein said salts are selected from salts K^+ , Na^+ , NH_4^+ , or containing R_3NH^+ and R_4N^+ -where wherein R is C_1 to C_4 alkyl.

Claim 7 (currently amended): A matched set of dyes-according to any of claims 1 to 6 selected from the group consisting of:

Set 1

 $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxohexyl})-2-[(1E,3E)-3-(1-\operatorname{ethyl}-3,3-\operatorname{dimethyl}-5-\operatorname{sulpho}-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{prop}-1-\operatorname{enyl}]-3,3-\operatorname{dimethyl}-3H-\operatorname{indolium} (\operatorname{Compound}\,\mathrm{I});$ and $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxohexyl})-3,3-\operatorname{dimethyl}-2-[(1E,3E,5E)-5-(1,3,3-\operatorname{trimethyl}-5-\operatorname{sulpho}-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{penta}-1,3-\operatorname{dienyl}]-3H-\operatorname{indolium} (\operatorname{Compound}\,\mathrm{II});$

Set 2

 $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxohexyl})-2-[(1E,3E)-3-(1-\operatorname{propyl}-3,3-\operatorname{dimethyl}-5-\operatorname{sulpho}-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{prop}-1-\operatorname{enyl}]-3,3-\operatorname{dimethyl}-3H-\operatorname{indolium}$ (Compound III); and $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxohexyl})-3,3-\operatorname{dimethyl}-2-[(1E,3E,5E)-5-(1-\operatorname{ethyl}-3,3-\operatorname{trimethyl}-5-\operatorname{sulpho}-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{penta}-1,3-\operatorname{dienyl}]-3H-\operatorname{indolium}$ (Compound IV);

Set 3

 $1-(6-\{[2-(2,5-\text{dioxo}-2,5-\text{dihydro}-1H-\text{pyrrol}-1-\text{yl})\text{ethyl}]\text{amino}\}$ -6-oxohexyl)-2-[(1E,3E)-3-(1-ethyl-3,3-dimethyl-5-sulpho-1,3-dihydro-2H-indol-2-ylidene)prop-1-enyl]-3,3-dimethyl-3H-indolium (Compound I); and $1-(5-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}$ -6-oxopentyl)-3,3-dimethyl-2-[(1E,3E,5E)-5-(1-ethyl-3,3-trimethyl-5-sulpho-1,3-dihydro-2*H*-indol-2-ylidene)penta-1,3-dienyl]-3*H*-indolium (Compound V);

<u>Set 4</u>

 $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxohexyl})-2-[(1E,3E)-3-(3,3-\operatorname{dimethyl}(1-\operatorname{sulpho}-\operatorname{butyl})-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{prop}-1-\operatorname{enyl}]-3,3-\operatorname{dimethyl}-3H-\operatorname{indolium} (\operatorname{Compound} VI); and \\1-(5-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}-6-\operatorname{oxopentyl})-3,3-\operatorname{dimethyl}-2-[(1E,3E,5E)-5-(3,3-\operatorname{dimethyl}-(1-\operatorname{sulpho}-\operatorname{butyl})-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{penta}-1,3-\operatorname{dienyl}]-3H-\operatorname{indolium} (\operatorname{Compound} VII).$

Set 5

 $1-(6-\{[3-(2,5-\text{dioxo}-2,5-\text{dihydro}-1$H-pyrrol-1-yl)propyl]amino}-6-\text{oxohexyl})-2-[(1$E,3$E)-3-(1-ethyl-3,3-dimethyl-5-sulpho-1,3-dihydro-2$H-indol-2-ylidene)prop-1-enyl]-3,3-dimethyl-3$H-indolium (Compound VIII); and <math display="block">1-(6-\{[2-(2,5-\text{dioxo}-2,5-\text{dihydro}-1$H-pyrrol-1-yl)ethyl]amino}-6-\text{oxohexyl})-3,3-dimethyl-2-[(1$E,3$E,5$E)-5-(1-ethyl-3,3-trimethyl-5-sulpho-1,3-dihydro-2$H-indol-2-ylidene)penta-1,3-dienyl]-3$H-indolium (Compound IV); and$

Set 6

 $1-(6-\{[3-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{propyl}]\operatorname{amino}\}$ -6-oxohexyl)-2- [(1*E*,3*E*)-3-(3,3-\text{dimethyl}(1-\text{sulpho}-\text{butyl})-1,3-\text{dihydro}-2*H*-\text{indol}-2-\text{ylidene})\text{prop}-1- enyl]-3,3-\text{dimethyl}-3*H*-\text{indol}\text{ium} (Compound IX); and

 $1-(6-\{[2-(2,5-\operatorname{dioxo}-2,5-\operatorname{dihydro}-1H-\operatorname{pyrrol}-1-\operatorname{yl})\operatorname{ethyl}]\operatorname{amino}\}$ - $6-\operatorname{oxohexyl})-3,3-\operatorname{dimethyl}-2-[(1E,3E,5E)-5-(3,3-\operatorname{dimethyl}-(1-\operatorname{sulpho-butyl})-1,3-\operatorname{dihydro}-2H-\operatorname{indol}-2-\operatorname{ylidene})\operatorname{penta}-1,3-\operatorname{dienyl}]-3H-\operatorname{indolium}$ (Compound X).

Claim 8 (currently amended): A method for labelling a mixture of proteins in a sample wherein each of said proteins contains one or more cysteine residues, said method comprising:

- i) adding to an aqueous liquid containing said sample a fluorescent dye selected from a matched set of fluorescent dyes wherein each said dye contains a target bonding group that is covalently reactive with said proteins; and
- ii) reacting said dye with said proteins so that said dye labels said proteins; eharacterised in that wherein all available cysteine residues in said proteins are labelled with said dye.

Claim 9 (currently amended): A method according to The method of claim 8, wherein said fluorescent dye is a cyanine dye.

Claim 10 (currently amended): A method according to The method of claim 9, wherein said cyanine dye contains a sulphonic acid or sulphonate group.

Claim 11 (currently amended): A method according to any of claims 8 to 10 The method of claim 8, wherein said target bonding group is selected from a the group consisting of maleimido group groups and an iodoacetamido groups group.

Claim 12 (currently amended): A method according to The method of claim 8, further comprising prior to step i), the a step of treating the protein with a reductant.

Claim 13 (currently amended): A method according to The method of claim 8, wherein said dye is used added in a range of 5 to 200nmol of dye per 50µg of protein.

Claim 14 (currently amended): A method according to The method of claim 8, wherein said labelling is performed at a pH in the range from 6.0 to 9.0.

Claim 15 (currently amended): A method for labelling one or more proteins in a sample, the method comprising:

i) adding to a liquid sample containing said one or more proteins a fluorescent dye selected from a matched set of fluorescent dyes each dye in said set having the formula (I):

$$Z^{1}$$
 R^{3}
 R^{1}
 R^{2}

(I)

wherein n is different for each said dye and is 1, 2, or 3;

 Z^1 and Z^2 independently represent the carbon atoms necessary to complete a phenyl or naphthyl ring system;

one of groups R^1 and R^2 is the group:

where Y is a target bonding group;

remaining group R^1 or R^2 is selected from $-(CH_2)_4-W$ or $-(CH_2)_r-H$; group R^3 is hydrogen, except when either R^1 or R^2 is $-(CH_2)_r-H$, in which case R^3 is W;

W is selected from sulphonic acid and sulphonate;

p is an integer from 3 to 6;

q is selected to be 2 or 3; and

r is an integer from 1 to 5;

and their salts thereof;

eharacterised in that and further wherein when n of two of said dyes differs by +1, one of p, q and r of said two dyes differs by -1; and

ii) incubating said dye with said sample under conditions suitable for labelling said one or more proteins.

Claim 16 (currently amended): A method according to The method of claim 15, wherein each of Z^1 and Z^2 represents the carbon atoms necessary to complete a phenyl ring system.

Claim 17 (currently amended): A method according to The method of claim 15, or claim 16 wherein:

n is selected to be 1 or 2;

p is selected to be 4 or 5;

q is-selected to be 2 or 3; and

r is-selected to be 1, 2 or 3.

Claim 18 (currently amended): A method according to any of claims 15 to 17-The method of claim 15, wherein said target bonding group Y is selected from a the group consisting of maleimido-group groups and an iodoacetamido groups group.

Claim 19 (currently amended): A kit comprising a matched set of fluorescent dyes comprising at least two different fluorescent dyes having the formula (I):

$$Z^1$$
 R^3
 R^3
 R^3
 R^3

wherein n is different for each said dye and is 1, 2, or 3;

 Z^1 and Z^2 independently represent the carbon atoms necessary to complete a phenyl or naphthyl ring system;

one of groups R¹ and R² is the group:

$$\begin{array}{c} O \\ \parallel \\ ---(CH_2)_p ----C - NH ---(CH_2)_q --- Y \end{array}$$

where Y is a target bonding group;

remaining group R^1 or R^2 is selected from $-(CH_2)_4-W$ or $-(CH_2)_r-H$; group R^3 is hydrogen, except when either R^1 or R^2 is $-(CH_2)_r-H$, in which case R^3 is W;

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W is selected from sulphonic acid and sulphonate;

p is an integer from 3 to 6;

q is selected to be 2 or 3; and

r is an integer from 1 to 5;

and their salts thereof;

characterised in that and further wherein when n of two of said dyes differs by +1, one

of p, q and r of said two dyes differs by -1.
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